# **Production of Quality Woody and Floral Crops Using Innovative Production Techniques**

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#### **Abstract**

For woody plants, accelerating seed germination by breaking seed dormancy and controlling flowering to bypass the juvenile period is a key to producing a quality plant in the shortest possible time. Germination of mature seeds of Styrax japonicus, as investigated by magnetic resonance imaging, was accelerated and germination percentage was increased with 1 to 2 months of warm stratification followed by 2 to 3 months of cold stratification. However, it required 4 to 5 years to produce quality plants when plants were started from seeds. High quality Styrax plants were produced in 2 years with vegetative propagation. However, over-wintering loss of rooted cuttings was a problem and should be minimized. With Ardisia crenata, flowering plants required 4 to 5 years when started from seeds but less than 2 years were necessary when propagated from rooted cuttings. Traditionally, bulbous crops are forced from large bulbs grown in the field one to two years. Dormancy must be broken to induce flowering with bulb cold/shoot photoperiod treatments in *Lilium* and high – warm – low temperature treatments in Ornithogalum. It would be desirable if the bulb production phase could be bypassed to shorten the total production time. Quality plants of L. longiflorum, L. × elegans, and interspecific hybrids of these two, LA hybrids, were produced from seeds, stem bulbils, or tissue cultured plants, respectively, in a year. Temperature manipulation at the time of floral induction was a key for successful forcing of Lilium. For Ornithogalum thyrsoides, selection of genotypes and combination of optimum concentrations of auxin and cytokinin for in vitro propagation using leaf explants was important to produce quality cut flowers and potted plants in less than a year.

#### INTRODUCTION

For many woody plants from temperate regions, warm stratification (WS) followed by cold stratification (CS) is required for seed germination. After germination, a juvenile phase exists during which plants are unable to flower. Therefore, to speed flowering, seed germination should be accelerated by treating mature seeds and the duration of the juvenile phase should be minimized and, if possible, bypassed.

Styrax japonicus Sieb. et. Zucc produces white flowers with yellow stamens from May to June. Warm stratification (WS) for 3 to 5 months followed by CS for 3 to 4 months has been recommended for breaking dormancy (Dirr, 1990; Kwon, 1995). One seedling flowered 4 months after germination, but it did not flower again in the following 3 years. Styrax may have a juvenile period longer than 4 years (Roh, unpublished data).

Proc. XXVII IHC-S5 Ornamentals, Now! Ed.-in-Chief: R.A. Criley Acta Hort. 766, ISHS 2008 Ardisia crenata exhibits a long juvenile period of up to 3 years (Roh, unpublished data) which prevents extensive investigation of the controlling mechanism of flowering. Rooted stem tip cuttings from mature plants could be a useful production technique to bypass the juvenile stage. Therefore, it is desirable to study whether commercially acceptable high quality plants can be produced in less than 2 years from vegetative propagation.

The growth and development cycles of many geophytes, including *Lilium* and *Ornithogalum*, from propagation to sale of marketable flowering plants are divided into three phases; bulb production phase, bulb programming phase, and greenhouse forcing. Traditionally, lilies and ornithogalum are forced using large bulbs produced from the field, two to three years for *L. longiflorum* (Blaney and Roberts, 1976), 1 to 2 years for *L.* × *elegans*, Asiatic hybrid lilies, and 2 to 3 years for Oriental hybrid lilies (Jeon et al., 2002). Tissue cultured propagules of *Ornithogalum thyrsoides* and *O. dubium* require approximately one year when raised in the greenhouse (Roh, unpublished data).

Lilies, except *L. longiflorum*, and ornithogalum need to be propagated vegetatively because of a segregation of progeny that results in non-uniform floral characters. For woody plants, propagation by cuttings or tissue culture needs to be investigated to avoid the long juvenile period.

The overall objectives in developing new production methods are to shorten the total production time while producing commercially acceptable plants, and to maximize the number of flower buds (Roh and Wilkins, 1977b) or increase the percentage of flowering plants. This review article reports the research results of seed germination and production of *Styrax japonicus* and *Ardisia crenata* from cuttings, and forcing *L. longiflorum* Easter lilies from seeds,  $L \times elegans$  Asiatic hybrid lilies from stem bulbils, and interspecific hybrid lilies between *L. longiflorum* and  $L \times elegans$  (LAIH), and *Ornithogalum thyrsoides* from tissue—cultured propagules to flower with a commercially acceptable quality.

### SEED GERMINATION, VEGETATIVE PROPAGATION, AND FLOWERING OF WOODY PLANTS

Information is lacking on how to shorten the juvenile period of woody plants and to produce quality flowering plants in short period of time. Further, the carry-over effect on growth and flowering of warm and cold stratification used to promote seed germination has not been reported. Acceleration of seed germination (Johnson and Roh, 2006) could possibly shorten the production time.

#### Styrax japonicus

Seedlings can not be forced to flower in less than 4 years, due to a long juvenile period (Roh, unpublished data). Rooted cuttings can produce acceptable size flowering plants in 3 years; however, many plants are lost over the winter (overwinter loss). Cuttings produced a healthy root system. The cause of this overwinter loss has not yet been determined. Plant production from seedlings is a possible alternative, if methods to shorten the duration of juvenility can be developed.

- 1. Seed Propagation. Warm stratification (WS) for 3 to 5 months followed by cold stratification (CS) for 3 to 4 months has been recommended (Dirr, 1990). However, one month of WS followed by 2 months of CS yielded higher than 80% germination (Roh et al., 2004) (Fig. 1). Seeds were mature when fruits were harvested in 12 weeks in 1999 and 16 weeks in 2000. Only one seedling flowered 6 months after germination, but failed to flower in the subsequent 4 years, indicating that the juvenile period could last longer than 4 years.

  2. Rooting of Cuttings to Bypass Juvenile Period. Styrax can be rooted easily by taking semi-hardwood cuttings between May and August. Although rooting hormones are
- semi-hardwood cuttings between May and August. Although rooting hormones are beneficial, they are not needed to get good rooting when propagated under mist (Fig. 2). The survival of the rooted cuttings is low when rooted cuttings overwinter in a greenhouse maintained at low temperature or in a cold frame. Root systems appear healthy when rooted cuttings start to wilt and all leaves collapse. Therefore, low overwinter survival is a

problem for development of a fast cropping system using stem tip cuttings, although some one-year old plants flowered (Fig. 3). The cause of the overwinter loss should be investigated. At least 3 years, with the first year for rooting of cuttings and over-wintering, the second year for increasing the size and shaping by pinching and pruning, and the third year to force to flower, could be necessary to produce marketable plants.

#### Ardisia crenata

Ardisia seeds are recalcitrant, and can not be stored like non-recalcitrant seeds. The juvenile period lasts 3 - 4 years. Rooting of cuttings was not influenced by the concentration of rooting hormone, but selected rooted cuttings were forced to flower and produced berries (Roh et al., 2005). About one-half of the rooted cuttings were uniform and were forced to flower.

- 1. Seed Propagation. Seeds are recalcitrant and can not be stored at room temperature longer than about 6 weeks (Roh, unpublished data). Therefore, seeds must be sown and germinated at  $20^{\circ}$ C as soon as harvested to achieve about 60 80% germination.
- **2. Rooting of Cuttings.** Ten cm long cuttings from 4-year-old stock plants grown in a greenhouse were collected and treated with 0, 1,000, 2,000, or 4,000 ppm of water-soluble potassium salt of IBA (IBA-K) solutions for 5 minutes and propagated under intermittent mist. Rooting percentage varied from 64% (control) to 82% (1,000 ppm IBA-K), however due to great variations ranging between 0 and 26 roots per cutting in a given treatment, means were similar regardless of the concentration of IBA-K solutions (Roh et al., 2005). The number of cuttings that can be collected from a 4-year-old non-branched stock plant is limited to less than 10, and this required many well branched stock plants.
- **3. Selecting Rooted Cuttings for Forcing.** Among 3 different plant types observed about one month after transplanting the rooted cuttings (Fig. 4), Type A plants producing only vegetative shoots were selected and forced. At transplanting, the number of vegetative shoots of Type A plants ranged from 5 to 10 and the length at transplanting ranged from 5 to 10 cm (data not presented). Type B plants produced reproductive shoots containing flower buds and vegetative shoots. Type C plants produced shoots with flower buds only, were the weakest in growth, and were unsuitable for forcing.
- **4. Criteria for Acceptable Plants.** Type B4 plants (produced berries from more than nine shoots on the upper part of the plant with the diameter of plants 1.5 times greater than that of pot) and type A3 plants (with vegetative shoots at the upper part of the plant and with more than five to eight reproductive shoots with berries beneath the vegetative shoots) are considered to have a high commercial value. Quality plants produced more than eight shoots with berries. Most shoots had more than five to eight berries per cluster; berry bearing shoots evenly spread over the pot rim; high commercial value (A3, Fig. 5) or those with more than nine shoots with berries and berries fully developed, the diameter of plants 1.5 times greater than that of pot (B4, Fig. 5).
- **5. Forcing.** When grown at 24D/21N°C, many A3, and B4 types produced berries only from old shoots while 14 plants produced berries from both old shoots and new shoots. The overall grades of plants grown in a GH at 21D/18N°C and 24D/21N°C were 3.9 and 4.1, respectively, which is significantly higher than the grade of plants grown at lower temperatures (Table 1).

#### DORMANCY, MATURITY, AND FLOWERING OF GEOPHYTES

#### Morphology and Physiology of the Bulb - Dormancy and Maturity

Lily and ornithogalum bulbs are composed of scales and a basal plate. The basal plate, a compressed stem, bears a shoot apex. Scales are modified swollen leaves containing food reserves (Rees, 1992). The size of the bulbs is largely determined by the number of scales and the degree of scale filling (bulb enlargement).

The site of dormancy in L. longiflorum and  $L \times elegans$  is considered to be the daughter scales (Roh and Wilkins, 1977a) and shoots, respectively, and a bulb cold-treatment is required to break dormancy and to induce early flowering. Although

unknown for ornithogalum, scales are believed to be the site of dormancy and a sequential treatment at warm and low temperatures is required to produce quality plants (Roh, unpublished data). Therefore, if scales are not present, dormancy breaking treatments may not be necessary.

### **Current Bulb Programming Methods and Their Implications for the Number of Flower Buds**

Lily bulbs are packed in wet peat moss and stored at 4.5 - 7.5°C for 6 weeks (Easter lily), 6 to 9 weeks (Asiatic hybrid lilies), and 9 weeks (Oriental lilies) (De Hertogh and Wilkins, 1971a, b; Roh and Wilkins, 1976; Roh, 1985). Bulb vernalization induces rapid and uniform shoot emergence and flowering, although the number of flower buds is reduced for Easter lily (Roh and Wilkins, 1976), but increased for Asiatic hybrid lily (Lee et al., 1996; Roh, 1985). Non-vernalized Asiatic and Oriental hybrid lilies failed to emerge or were delayed, respectively, and plants subsequently fail to reach anthesis. Therefore, for these two lilies, a minimum bulb vernalization treatment should be given for flowering.

A long-day photoperiod treatment given to *L. longiflorum* accelerates shoot emergence, but also reduces the number of flowers (Roh and Wilkins, 1977b). In Asiatic hybrid lilies, a shoot photoperiod treatment did not accelerate flowering (Roh, 1985). For *L. speciosum*, a long-day photoperiod is effective for accelerating flowering when bulbs do not receive enough vernalization treatment (Ohkawa, 1977).

Conventional 6 weeks of continuous vernalization or 30 days of continuous long day photoperiod treatments reduce the number of flower buds, even though flowering is accelerated. Therefore, to avoid reduced flower numbers it was suggested to divide 6 weeks of bulb vernalization into 2 periods of 3 weeks vernalization which are separated by non-vernalizing temperatures (Table 2) (Roh and Wilkins, 1976). The approach in developing new production techniques, particularly using small size propagules like seedlings, bulbils, and tissue cultured propagules, should modify bulb programming methods to increase the potential number of flowers.

Ornithogalum bulbs are stored dry, similar to tulip, at room temperatures or at the respective temperatures to induce flower bud formation. The optimum level of humidity which may affect the growth of initiated root primordial during storage has not been investigated.

#### **Approaches to Develop New Production Technologies**

To develop a new production technique, attention should be given to the shoot apex where growth and flowering eventually takes place after the requirement of bulb cold-treatment is satisfied (Roh and Lawson, 1992). Since the number of flowers resulting from an increased size of shoot apex increases, it is possible to produce flowering plants starting from small bulblets or seedlings (Table 3).

#### **New Production Technologies**

Three process patents to produce lilies have been granted and brief descriptions - duration of certain important growth and development events of the technologies - were summarized (Roh, 2002). Since the other two methods (Oglevee and Tammen, 1986; Tammen, 1991) have not been evaluated extensively, US Patent no. 5,138,794 "Method for producing *Lilium* species" by Roh (1992) will be covered briefly in this review article.

#### APPLICATION OF NEW PRODUCTION TECHNOLOGIES - LILIUM

Cultural information is described briefly here for producing commercially acceptable quality cut flowers or potted plants of the major three types of lilies. More details can be found in the previous articles (Roh et al., 1996; Roh, 2002).

#### L. × elegans from Stem Bulbils

A method for producing *Lilium elegans* from bulbils as propagules is briefly described (Roh, 1992).

- **1. Bulbil Harvest and Temperature Treatments.** Approximately 45 60 days after flowering, 400 to 500 mg bulbils are harvested, packed in moist peat moss, and then receive 20 days of 5 7.5°C followed by 7 to 14 days of 10 12.5°C followed by 20 days of 5 7.5°C, and then planted in 10-cm pots (Fig. 6).
- **2. Temperature Control after Potting.** Temperatures are maintained at 25 32°C during the day and 21 26°C at night or until no new scaly leaves are formed (Fig. 7).
- 3. Low Temperature and Long-Day Photoperiod Treatment. When short and wide scaly leaves appear as the first sign of shoot formation, night temperature is dropped to 10 12.5°C with the day temperature maintained at 15.5 21°C for 40 to 60 days.
- **4. Transplanting into a Final Size Pot or Bench.** Plants are transplanted into either a 12.5 cm pot for pot culture or into a bench for cut flower production. Temperatures are raised to 21°C/15.5°C, day/night, until flowering. Because temperatures before shoot emergence are maintained at 10 12.5°C for 40 60 days, it may not be necessary to maintain these low temperatures after shoot emergence.

#### L. longiflorum from Seeds

In the following section the results of Easter lily experiments designed to produce commercially acceptable quality plants from seed are presented. Some relevant information is presented here. Advanced breeding efforts have been made recently (Song et al., 2005).

- **1. Seed Germination.** *Lilium longiflorum* seeds germinate best at 16°C within 30 days of maturity.
- **2. Selection of Seedlings and Stock-Plant Preparation for Seed Production.** Seedlings showing leaf scorch on the cotyledon or true leaves were discarded during the initial seedling selection and stock-plant culture for seed production. All selected seedlings were grown in a 21/16°C greenhouse until November/December and those that failed to show emerging shoots were also discarded.
- **3. Flowering.** Plants become uniform in growth and flowering, and growing from seeds is economically feasible. Plants that were exposed to 26/24°C produced an average of 4.6 flowers, with 60% of plants producing 5 or 6 flowers. However, several plants produced flower buds with an abnormal shape. Formation of normal and abnormal flowers is another concern which may be avoided by selecting proper stock plants for seed production (Table 4).

## L. longiflorum × L. x elegans (LA) Hybrid Lilies (LAIH) from Tissue Cultured Propagules

This has been reported in detail by Roh et al. (1996a, b), however, production technologies are briefly described here. There are many LA hybrids available on the market; however, some of them are hybrids between L. × *formolongi* and L. × *elegans*. Therefore, LAIH hybrids were created and evaluated. Preliminary evaluation of Ornipet lilies (a hybrid between Oriental lily and trumpet lily) was not conclusive.

- **1. Propagation and Culture of Selected LAIH Hybrids.** Five selected LAIH offspring ('A 8', 'A 159', 'A 168', 'A 238', and 'A 274') and the two parents ('Nellie White' and 'D') were tissue-culture propagated on a Murashige and Skoog (MS) medium supplemented with 3 mg/L BA and 0.5 mg/L NAA.
- **2. Growth and Flowering.** The days to flower of all LAIH plants did not differ from that of the two parents and ranged from 116 to 127 days after emergence. 'Nellie White' produced 4.5 primary, 1.5 secondary, and 0.7 tertiary flower buds. This result clearly indicates that large-size bulbs are not required to produce commercially acceptable cut flowers or potted plants (Roh and Lawson, 1992).

#### APPLICATION OF NEW PRODUCTION TECHNOLOGIES – ORNITHOGALUM

In *O. thyrsoides*, dormancy may not be too deep and further dormancy may not be an issue if tissue cultured propagules can be forced directly without any bulb temperature treatments. However, *O. dubium* bulbs enter dormancy and dormancy should be released (Roh and Joung, 2004). Information on flower induction of *Ornithogalum* as affected by

long day photoperiod is not available; however, preliminary tests indicated that flowering of *Ornithogalum* is mainly affected by temperature.

**Forcing Starting from Bulb** 

The possibility to produce quality cut flowers and potted plants starting from tissue cultured propagules has not been investigated in detail. *Ornithogalum thyrsoides*, which has a shallow dormancy, was investigated extensively at the Floral and Nursery Plants Research Unit. *O. dubium* hybrids which have a relatively deeper dormancy were not as thoroughly investigated. *Ornithogalum thyrsoides* may not enter into dormancy and new leaves continue to form if plants are watered continuously at 16°C (Halevy, 1990). Dormancy of *O. dubium* bulbs must be broken by high temperatures to induce flowering (Fig. 14) (Roh and Joung, 2004). To shorten the total production time one should consider bypassing a bulb production phase. Optimum temperature to break dormancy and to induce floral initiation and differentiation is around 25°C. Once dormancy is broken, floral organs are formed in the center of the bulb (Table 5).

Flowering was not affected by bulb maturity when sequential bulb temperature programming treatments were given. Immature bulbs, treated at 30°C for 3 weeks (30°C/3 wk), flowered earlier than mature bulbs and flowering percentage was higher. Immature bulbs that received 30°C/3 wk followed by 10°C/2 wk flowered significantly later than mature bulbs. However, when bulbs received 30°C/6 wk, regardless whether 30°C treatment was given continuously for 6 weeks or 2 weeks of 10°C was given in the middle of 30°C (30°C/2 wk - 10°C/2 wk - 30°C/3 wk) treatment, there was no significant difference in the days to flower and flowering percentage (Table 6). Therefore, in *O. thyrsoides*, bulb maturity can be induced by temperature treatments as in *L. longiflorum* (Roh and Wilkins, 1977a) and bulbs can be harvested when leaves were still growing. However, this early harvest can not shorten the production time greatly.

#### Forcing, when Starting from Tissue Cultured Propagules

Tissue cultured propagules may not need any treatment if leaves lacking swollen leaf bases are allowed to develop leaves and a scape continuously without initiating bulb formation. Leaf base may not be enlarged, and this eliminates the site for a possible presence of dormancy factors. More than 40 genotypes were evaluated for the feasibility study and here is some brief information on the production of commercially acceptable cut flowers from tissue cultured propagules.

#### **Tissue Culture Propagation**

Explants are cultured on a MS medium supplemented with 0.5 mg/L NAA and 3 mg/L BA. Clumps were divided into single plantlets and grown in a greenhouse maintained at 16.5/16°C.

#### **Greenhouse Culture**

Plantlets were planted in 5-cm square pots first filled with soilless medium and then transplanted in 5-cm deep soilless medium in trays or plantlets are planted directly into trays filled with soilless medium and grown until flowering. Flowering date was recorded when two florets opened and other data were collected.

#### **Flowering**

Plants take about 90 days to flower and the stem length varies from 80 - 100 cm. Stem are very strong and support more than 200 florets per scape, depending on the forcing season.

#### PROSPECTS AND LIMITATIONS

The cause of overwinter loss in *Styrax* cuttings should be investigated and solved to make rooting of cuttings feasible for commercial production. For *Ardisia*, improved rooting percentage and the percentage of rooted cuttings that can be forced should be understood.

Commercial-scale production should be attempted. The market share and impact of lilies produced using these techniques either as potted plants or cut flowers will be low at the beginning. But these techniques may open the door to the production of lilies under a controlled-environment system. Although *L. longiflorum*, *L.* × *elegans*, and LAIH hybrid lilies can be produced utilizing these new methods by eliminating bulb-production phase, the prospect of utilizing these techniques are most promising with *L. longiflorum* and possibly with LAIH hybrids.

For *Ornithogalum*, quality cut flowers and potted plants were produced. However, optimum greenhouse forcing temperatures should be investigated, since only one forcing temperature was tested. Forcing temperatures even 2 to 5°C higher than 16.5/16°C seem to

be too high. This will limit the year-round production.

These technologies have been shown to be practical and are available, although commercial and large-scale production trials must still be performed by growers. Although it is feasible to produce quality products in less than a year, this may be considered too long to grow and force. Plug production technologies should be employed so that propagules from seeds, bulbils, and tissue culture can be grown for 6 to 7 months by plug producing specialists, and those can be supplied to growers to finish the crop in 4 to 5 month.

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#### **Tables**

Table 1. Development of *Ardisia crenata* from rooted cuttings as influenced by greenhouse forcing temperatures (Roh et al., 2005, modified).

GH temp.	Number of plant	Overall Grade <sup>2</sup>		
(°C)		$\underline{\text{Type A2} + \text{A3} + \text{B4}}$		
	Old shoots	New shoots <sup>1</sup>		
15/12	16	8	1.4	
18/15	9	19	2.3	
21/18	4	24	3.9	
24/21	1	26	4.1	
Total	30	77	-	
hsd at 5%	-	-	0.7	

<sup>&</sup>lt;sup>1</sup> Both old and new berry-bearing shoots are included in counts of fruiting plants.

<sup>2</sup> Grade scale: 1 - unacceptable; 2 - unacceptable; 3 - marginally acceptable;

4 - acceptable; 5 - superior.

Table 2. The effect of continuous and interrupted bulb vernalization and shoot photoperiod treatment on growth and flowering of *L. longiflorum*, 'Nellie White' and 'Ace' (modified after Roh and Wilkins, 1976).

Treatment	No. of days to flower		Number of flowers			
(vernalization or photoperiod)			tota	total		primary
	'Ace'	'NW'	'Ace'	'NW'	'Ace'	'NW'
Bulb vernalization						
15.6°C/8 wk	189	193	9.6	13.0	5.4	7.4
4.4°C/6 wk - 15.6°C/2 wk	175	176	6.8	7.0	4.6	5.0
15.6°C/3 wk - 4.4°C/5 wk	159	145	6.4	7.6	4.6	5.0
4.4°C/1 wk - 15.6°C/3 wk - 4.4°C/4 wk	178	181	6.8	9.8	5.0	5.4
Shoot photoperiod						
Short day photoperiod (ND)	170	167	11.0	15.4	6.0	9.4
$30  \text{LD} - 30  \text{ND}^{1}$	119	126	6.8	7.4	5.0	5.4
30 ND - 30 LD	117	120	6.8	7.2	4.6	5.0
<u>10 LD - 20 ND - 20 LD</u>	124	127	8.6	9.0	4.6	4.8

<sup>&</sup>lt;sup>1</sup> Long day was given as 5 hours of night interruption from 10 pm to 3 am.

Table 3. Propagation methods of various *Lilium* species and hybrids that can be used as propagules to develop new production technologies.

Species/ hybrids	Seed	Bulblets Tissue culture Leaf cutting		Scaling	Bulbil
L. longiflorum	no/yes <sup>1</sup> yes no no	yes	yes	yes	no
L. × formolongi		no	no	yes	no
L. × elegans		yes	no/yes	yes	yes
LAIH hybrids		yes	no	yes	yes/no

<sup>&</sup>lt;sup>1</sup> Young propagules can be used (yes) or can not be used (no) in developing new production technologies.

Table 4. Flowering of two *L. longiflorum* 'Nellie White'  $\times$  ('Ace'  $\times$  'Nellie White') hybrids as influenced by forcing temperatures.

Temp	No. of days to flower from		No. of	Flower shape <sup>1</sup>		Plant height	
$(^{\circ}C)^{T}$	Sowing	Treatment	flower	Α	Ň	(cm)	
'Nellie W	/hite' × ('Ace' ×						
18/16	377	89	4.6	2	11	37	
22/20	366	78	4.2	1	11	33	
26/24	359	71	4.0	3	11	31	

<sup>&</sup>lt;sup>1</sup> Flower bud shape was abnormal (A) or normal (N).

Table 5. Growth and flowering of *O. dubium* as influenced by bulb storage temperature.

Storage	No. of plants		Number of days to					
Temp (°C)	Flowered <sup>1</sup>	Leaf emerged	Flowering	$FL - LE^2$				
Ornithogalum dubium (1999)								
10	0	-	=	-				
15	2	58	183	125				
20	2	44	198	124				
25	14	4	122	118				
30	17	4	124	120				
_ 35	11	36	151	115				
Level of significance <sup>3</sup>								
Linear		ns	ns	ns				
Quadratic		**	**	**				

<sup>&</sup>lt;sup>1</sup> Flowering of the primary and visible/presence of the secondary inflorescence. Each treatment has 22

Table 6. Effect of bulb maturity and bulb storage temperatures on growth and flowering of Ornithogalum, 'Chesapeake Starlight' (Roh and Hong, unpublished data).

Date of potting	Bulb maturity	EM	o. of da FL		FL L %
Apr. 13	immature	103	293	190	48
Apr. 13 July 8	mature immature	127 9	296 161	169 152	23 100
July 8 Aug. 5	mature immature	9 2	153 115	144 113	100 100 100
	Apr. 13 Apr. 13 July 8 July 8	Apr. 13 immature Apr. 13 mature July 8 immature July 8 mature July 8 mature Aug. 5 immature	potting maturity EM  Apr. 13 immature 103 Apr. 13 mature 127 July 8 immature 9 July 8 mature 9 Aug. 5 immature 2	potting         maturity         EM         FL           Apr. 13         immature         103         293           Apr. 13         mature         127         296           July 8         immature         9         161           July 8         mature         9         153           Aug. 5         immature         2         115	potting         maturity         EM         FL         EM-F           Apr. 13         immature         103         293         190           Apr. 13         mature         127         296         169           July 8         immature         9         161         152           July 8         mature         9         153         144           Aug. 5         immature         2         115         113

 $<sup>^{1}</sup>$  No. of days to emergence (EM) and flowering (FL) was counted from the dates of potting.  $^{2}$  Root primordia visible on June 22, and bulbs were stored at  $10^{\circ}$ C.

bulbs.

<sup>2</sup> LE: Leaf emergence; FL: Flowering, FL - LE: Number of days from leaf emergence to flowering.

<sup>3</sup> ns, \*, \*\*; non-significant, significant at 5%, 1% level. Data from bulbs treated at 10°C was excluded from analysis.

### **Figures**

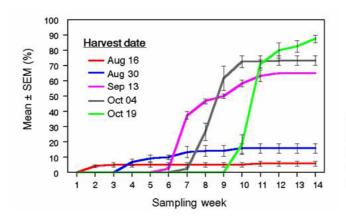


Fig. 1. Styrax seed germination as influenced by harvest dates. Refer to Roh and Bentz (2003) and Roh et al. (2005) articles for details of seed treatments.



Fig. 2. Well rooted Styrax japonicus cuttings.



Fig. 3. One year old Styrax japonicus cutting at flowering.

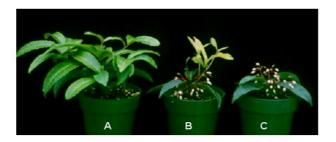


Fig. 4. Three types of *Ardisia crenata* after transplanting of rooted cuttings. Type A plant produced only vegetative shoots, type B plant produced shoots with flowers and also with a few vegetative shoots, and type C plant produced shoots with flower buds only.



Fig. 5. Appearance of *Ardisia crenata* produced from cuttings. Plant A3 formed berries beneath the vegetative shoots and plant B4 formed berries without the vegetative shoots.



Fig. 6. Stem bulbils of Asiatic hybrid lilies,  $L. \times elegans$ .



Fig. 7. Formation of scale leaves and small bulbs around October.



Fig. 8. L. longiflorum seedlings in 10 cm pot.



Fig. 9. Selected seedling at transplanting. Contractile root (arrow) is just formed.



Fig. 10. L. longiflorum grown from seeds at flowering.

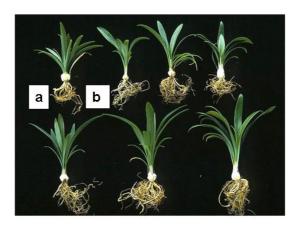


Fig. 11. Tissue cultured propagules of L. longiflorum (a),  $L \times elegans$  (b), and 5 interspecific hybrids (LAIH) (not indicated). Hybrid vigor is noticed in some LAIH hybrids.



Fig. 12.  $L. \times elegans$ , LAIH, and L. longiflorum (from left to right) forced from tissue cultured propagules at visible buds stage. Scaly leaves are still attached in L. longiflorum and LAIH hybrid.



Fig. 13. L. × *elegans*, LAIH, and L. *longiflorum* (from left to right) forced from tissue cultured propagules at flowering.

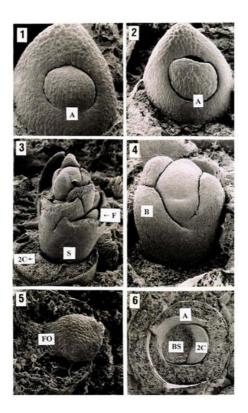


Fig. 14. Inflorescence initiation and development of *Ornithogalum* hybrid as affected by bud storage temperatures; 10°C (1), 15°C (2), 20°C (3), 25°C (4), and 30°C (5). Scape of the secondary inflorescence is visible (2C) (3, 6). (For label details, refer to Roh and Joung, 2004).



Fig. 15. Selection of genotype to study the feasibility of producing *O. thyrsoides* starting from tissue culture propagules.



Fig. 16. Clumps of tissue cultured plantlets of *O. thyrsoides*.



Fig. 17. Greenhouse production of *O. thyrsoides* starting from tissue cultured propagules. Separated plantlets are planted in 5 cm pots, and then transplanted in tray with 5-cm deep soilless growing medium.